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HELVETICA CHIMICA ACTA, vol. 60, Fasc. 1, no. 24, 1977, pages 211-14, Basel (CH); B. SCHIRCKS et al.: "24. Über pterinchemie. Eine neue, regiospezifische Synthese von L-Biopterin"

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(56) References cited:

HELVETICA CHIMICA ACTA, vol. 61, Fasc. 7, no. 257, 1978, pages 2731-38, Basel (CH); B. SCHIRCKS et al.: "257. Über Pterinchemie: Herstellung von (6R,S)-5,6,7,8-Tetrahydro-L-biopterin, 7,8-Dihydro-L-biopterin, L-Sepiapterin, Deoxysepiapterin, (6R,S)-5,6-Dihydrodeoxysepiapterin und 2'-Deoxybiopterin)"

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Courier Press, Leamington Spa, England.

Description

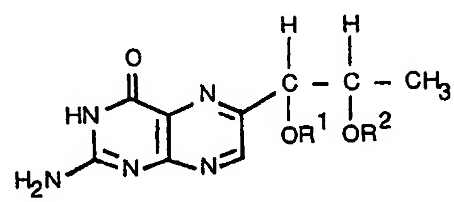
It has been found recently that (6R,S)-5,6,7,8-tetrahydro-L-biopterin and 7,8-dihydro-L-biopterin can be successfully used for a treatment of patients with a typical phenylketonurea (hereinafter referred to as PKU) or with dihydropterin-reductase deficiency [A. B. Schircks, M. Viscontini and J. Schaub, Lancet, 1979, 131; H.-Ch. Curtius, A. Niederwieser, M. Viscontini, A. Otten, J. Schaub, S. Scheibenreiter and H. Schmidt, Clin. Chim. Acta. *93*, 251 (1979)].

Though both compounds can conduct an enzymatic hydroxylation of L-tryptophane and L-tyrosine to 5-hydroxytryptophane and DOPA, respectively, in the brain, they have difficulties to cross the blood brain barrier. Therefore neurotransmitter precursors must be given with those compounds during the treatment of both deficiency-diseases. From US—A—3,505,329 it is known to prepare biopterin via 5-deoxy-L-arabinose as an intermediate and from CH—A—500,999 it is known to prepare neopterin and monopterin starting from a sugar hydrazone.

The present invention has been completed on the basis of the fact that lipophile substances can more readily cross the brain barrier than compounds which possess a greater polarity as (6R,S)-tetrahydro-L-biopterin with its amphoteric nucleus and its free sugar chain.

An object of the present invention is to provide a process for preparing 1',2'-diacyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin having a low polarity and the same effect as (6R,S)-5,6,7,8-tetrahydro-L-biopterin in treatment of atypical PKU and dihydropterin-reductase deficiency without neurotransmitter precursors.

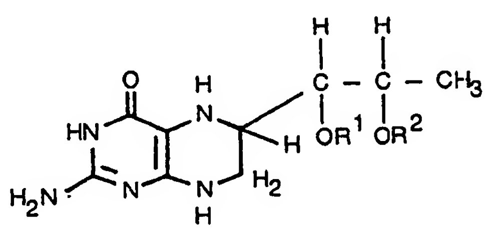
In accordance with the present invention, there is provided a process for preparing a 1',2'-diacyl-L-biopterin having the general formula (II):



(II)

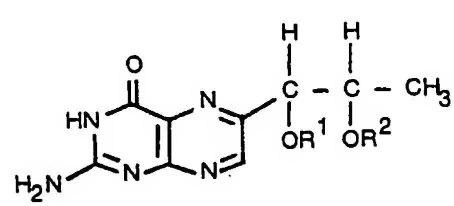
wherein R^1 and R^2 are the same or different and each is an acyl group, by converting L-rhamnose to an acyl derivative of 5-deoxy-L-arabinose-phenylhydrazone through a 1,1-dialkylsulfonyl-L-rhamnose, reacting the obtained acyl derivative with 2,4,5-triamino-6-hydroxy-pyrimidine and then oxidizing the product with an oxidizing agent, which is characterised in that the reactions from the 1,1-dialkylsulfonyl-L-rhamnose to the 1',2'-diacyl-L-biopterin are carried out without isolating the intermediate products.

Furthermore, there is provided a process for preparing a 1',2'-diacyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin having the general formula (I):



(I)

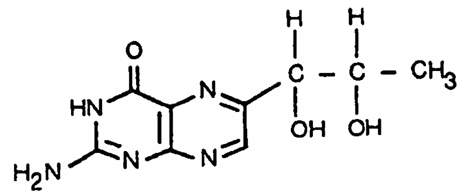
wherein R^1 and R^2 are identified as above which comprises converting L-rhamnose to an acyl derivative of 5-deoxy-L-arabinose-phenylhydrazone through a 1,1-dialkylsulfonyl-L-rhamnose, reacting the obtained acyl derivative with 2,4,5-triamino-6-hydroxypyrimidine and then oxidizing with an oxidizing agent to obtain a 1',2'-diacyl-L-biopterin having the general formula (II):



(II)

wherein R^1 and R^2 are as defined above, which is characterised in that the reactions from the 1,1-dialkylsulfonyl-L-rhamnose to the 1',2'-diacyl-L-biopterin are carried out without isolating the intermediate products and that the 1',2'-diacyl-L-biopterin is catalytically hydrogenated in a solvent in the presence of a hydrogenation catalyst.

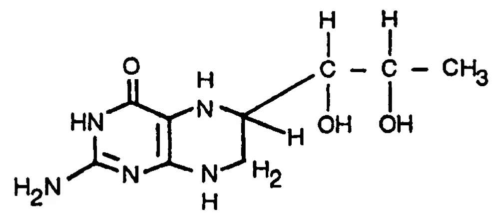
Further there is provided a process for preparing biopterin having the formula (II'):



(II')

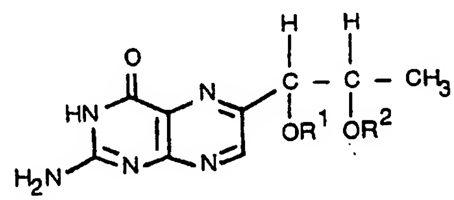
by converting L-rhamnose to an acyl derivative of 5-deoxy-L-arabinose-phenylhydrazone through a 1,1-dialkylsulfonyl-L-rhamnose, reacting the obtained acyl derivative with 2,4,5-triamino-6-hydroxy-pyrimidine and then oxidizing with an oxidizing agent and deacylating the resultant 1',2'-diacyl-L-biopterin, which is characterised by carrying out the reactions from the 1,1-dialkylsulfonyl-L-rhamnose to the L-biopterin without isolating the intermediate products.

Further there is provided a process for preparing a (6R,S)-5,6,7,8-tetrahydro-L-biopterin having the formula (I'):



(I')

which comprises converting L-rhamnose to an acyl derivative of 5-deoxy-L-arabinose-phenylhydrazone through a 1',1-dialkylsulfonyl-L-rhamnose, reacting the obtained acyl derivative with 2,4,5-triamino-6-hydroxy-pyrimidine and then oxidizing with an oxidizing agent to obtain a 1',2'-diacyl-L-biopterin having the general formula (II):



(II)

wherein R^1 and R^2 are as defined above, and deacylating the resultant 1',2'-diacyl-L-biopterin without isolating the intermediate products in the course of reactive from the 1,1-dialkylsulfonyl-L-rhamnose to the L-biopterin, and catalytically hydrogenating the L-biopterin in a solvent in the presence of a catalyst.

The compound having the general formula (I) can be used for treatment of atypical PKU or dihydropterin-reductase deficiency and can readily cross the blood brain barrier without neurotransmitter precursors.

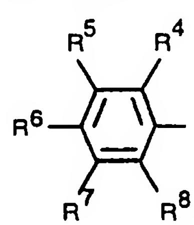
Fig. 1 and Fig. 2 represent, respectively, ^1H—NMR-spectrum chart and ^{13}C—NMR-spectrum chart of 1',2'-diacetyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin.

Fig. 3 represents ^1H—NMR-spectrum chart of 1',2'-dibutyryl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin.

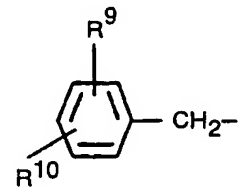
Fig. 4 represents ^1H—NMR-spectrum chart of 1',2'-dibutyryl-L-biopterin.

The acyl groups represented by R^1 and R^2 in the general formulas (I) and (II) are protective groups of the hydroxyl groups of (6R,S)-5,6,7,8-tetrahydro-L-biopterin.

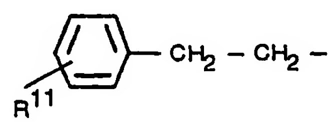
The acyl group has preferably 1 to 10 carbon atoms, in particular 3 to 10 carbon atoms. Preferable acyl group is represented by the general formula R^3CO— wherein R^3 is hydrogen or a hydrocarbon residue having 1 or more carbon atoms, in particular 2 to 9 carbon atoms. Preferable examples of the hydrocarbon residue represented by R^3 are, for instance, a linear or branched alkyl group having 1 or more carbon atoms, preferably 2 to 9 carbon atoms, which is either saturated or unsaturated; a substituted or unsubstituted phenyl group represented by the general formula



wherein R^4, R^5, R^6, R^7 and R^8 are hydrogen or a linear or branched alkyl group in which the total of the carbon atoms thereof is preferably not more than 3; a substituted or unsubstituted benzyl group represented by the general formula



wherein R^9 and R^{10} are hydrogen, methyl group or ethyl group in which the total of the carbon atoms thereof is preferably not more than 2; and a substituted or unsubstituted arylalkyl group represented by the general formula



wherein R^{11} is hydrogen or methyl group. Among the above acyl groups, formyl group, acetyl group, propionyl group, butyryl group, isobutyryl group, valeryl group, isovaleryl group and benzoyl group are most preferable. It is preferable that R^1 and R^2 are the same.

The compound of the general formula (I) has two diastereomers, i.e. 1',2'-diacyl-(6R)-5,6,7,8-tetrahydro-L-biopterin and 1',2'-diacyl-(6S)-5,6,7,8-tetrahydro-L-biopterin which are diastereomeric at the 6 position. The compound of the present invention includes the two diastereomers and a mixture thereof.

The compound of the present invention can be readily prepared by means of catalytic hydrogenation of the compound of the general formula (II) in a suitable solvent in the presence of a hydrogenation catalyst.

Examples of the catalyst are, for instance, Pt, Ni, Cr, Pd and Rh.

Examples of the solvent are, for instance, a solvent in which the compound (II) is soluble, such as trifluoroacetic acid, methanol, ethanol, propanol (1) and propanol (2), a conc. HCl, an acidic water, a basic water, or the like and a solvent in which the compound (II) is insoluble, but the compound (I) is soluble, such as acetic acid.

When the solvent which dissolves the compound (II) is employed, the process of the present invention can be carried out in a similar manner to the known process for preparing tetrahydro-L-biopterin by hydrogenating L-biopterin in trifluoroacetic acid in the presence of Pt [Bernard Schircks, Jost H. Bieri and Max Viscontini, Helvetica Chimica Acta, *61(7)*, 2731 (1978)].

In case where the compound (I) is used as an active ingredient of a pharmaceutical composition, it is not preferable that any solvent which is not pharmacologically acceptable remains. In such view point, the employment of acetic acid as a solvent is practically useful. The catalytical hydrogenation of L-biopterin can also be carried out by using acetic acid as a solvent.

As a result of the catalytical hydrogenation, a mixture of 1',2'-diacyl-(6R)-5,6,7,8-tetrahydro-L-biopterin and 1',2'-diacyl-(6S)-5,6,7,8-tetrahydro-L-biopterin is, in general, obtained in a proportion of about 1:1. The mixture can be resolved, for example, by means of high pressure liquid chromatography which is adapted to a resolution of (6R,S)-tetrahydro-L-biopterin [J. Biol. Chem., *253,* 1593 (1978)].

The compound (II) can be prepared by a similar process to the process for preparing 1',2'-diacetyl-L-biopterin [Bernard Schircks, Jost H. Bieri and Max Viscontini, Helvetica Chimica Acta, *60(1),* 211 (1977)]. According to the process, L-rhamnose hydrate is reacted with ethanethiol. The obtained L-rhamnose-diethylmercaptal is converted to 5-deoxy-L-arabinose through 1,1-diethylsulfonyl-L-rhamnose. 5-Deoxy-L-arabinose is reacted with phenylhydrazine to obtain 5-deoxy-L-arabinose-phenylhydrazone. 5-Deoxy-L-arabinose-phenylhydrazone is reacted with an acylating agent. The obtained 2,3,4-triacyl-5-deoxy-L-arabinose-phenylhydrazone is reacted with 2,4,5-triamino-6-hydroxy-pyrimidine dihydrochloride, and then oxidized with an oxidizing agent such as iodine to obtain a 1',2'-diacyl-L-biopterin. L-Biopterin can be obtained by deacylation of the 1',2'-diacyl-L-biopterin.

The above-mentioned process, however, is not suitable for industrial processes because large amount of solvent is required and the total yield of 1',2'-diacyl-L-biopterin is low due to its complicated procedures.

The inventor has found the fact that when the course of reaction from the 1,1-dialkylsulfonyl-L-rhamnose to the 1',2'-diacyl-L-biopterin, if necessary, to L-biopterin are carried out without isolating the intermediate products, that is to say, in one pot synthesis, it is possible to improve the total yield, to reduce the amount of the solvent and to simplify the procedures. The improved process of the present invention is quite suitable for industrial processes. As a mercaptal, methylmercaptal and propanethiol may be employed in addition to ethanthiol. In case of employing methylmercaptal and propanethiol, the respective starting compound of the improved process is 1,1-dimethylsulfonyl-L-rhamnose and 1,1-dipropylsulfonyl-L-rhamnose.

The improved process of the present invention can also be adapted to a production of L-biopterin and to productions of monapterins and neopterins which have different side chains at the 6 position.

Among the compounds (II), the 1',2'-diacyl-L-biopterin having the acyl group of 3 or more carbon atoms is a novel compound.

The compound (I) can be isolated from a reaction mixture in the form of an inorganic salt such as a hydrochloride, a sulfate or a phosphate, an organic salt such as an acetate, an oxalate, or a complex salt.

The present invention is more specifically described and explained by means of the following Examples.

Example 1

A) One pot synthesis of 1',2'-dibutyryl-L-biopterin

A suspension of 14 g (42.1 mmol) of diethylsulfonyl-L-rhamnose in 120 ml of water was treated with 4N NH_4OH while stirring until the pH of the solution.became 9 to 10. After 14 hours standing with stirring from time to time, the precipitate of diethylsulfonyl-methane was filtered off and the filtrate was dried in vacuo at 40°C. The residue was dissolved in 80 ml of absolute methanol. After adding 5 g (46 mmol) of pure phenylhydrazine, the solution was kept at room temperature for 1 hour and then dried in vacuo at 40°C. The residue was washed two or three times with ether (50 ml each time) and dried. The dried residue was dissolved in 35 ml of pyridine and the solution was cooled. To the ice-cold solution of 0—5°C 35 ml of butyric anhydride were slowly added. After adding completely, the reaction mixture was allowed to stand in the ice-bath for 10 min and then it was kept at room temperature for 5 hours. After 200 ml of methanol were added to the solution, the solution was kept at room temperature for 10—15 hours (overnight). A solution of 1.0 g of sodium dithionite ($Na_2S_2O_4$) and 12.5 g of sodium acetate·$3H_2O$ dissolved in 300 ml of water, and a suspension of 12.0 g of 6-hydroxy-2,4,5-triamino-pyrimidine sulfate·H_2O in 500 ml of water were then added successively to the methanol/pyridine solution. The whole reaction mixture was closed under nitrogen and stirred at 35—40°C for 20 hours to give an homogeneous, reddish brown solution. In the resulting solution the obtained tetrahydrobiopterin derivative was oxidized by adding 25 g of iodine dissolved in 300 ml of methanol. The excess of iodine, if any, was removed with sodium thiosulfate. If no excess was present, a small quantity of iodine solution was added in order to complete the oxidation of the tetrahydrobiopterin derivative. During the oxidation a fine brown crystalline precipitate was obtained.

The resulting oxidized suspension was concentrated to 50 ml in vacuo at 40°C and filtered. The insoluble fraction was washed with water, 40 ml of cold ethanol and finally with ether. The residue was then recrystallized from hot ethanol with active charcoal and the crystal was filtered while heating.

After cooling, yellowish precipitate of 1',2'-dibutyryl-L-biopterin was obtained in the filtrate. It was filtered, washed with ether and dried to give 10.3 g of 1',2'-dibutyryl-L-biopterin. Yield: 65%.

The obtained 1',2'-dibutyryl-L-biopterin was analyzed by means of several analytical methods. The ^1H—NMR-spectrum chart is shown in Fig. 4.

Rf (3% NH_4Cl/H_2O, on cellulose) = 0.36

$[\alpha]^{220}_{589} = -74.3 \pm 3°$ (C = 0.8, 1 N HCl).

The optical rotation increased with the time, perhaps because the butyrate ester is hydrolyzed in an acidic solution.

^1H—NMR-spectrum analysis (δ: p.p.m.): (90 MHz, in DCl) 933 (s, H—C (7); 6.5 (d, H—C (1')); 5—6 (m, H_2O, H—C (2')); 3—2.66 (m, $2xH_2C$—(CH_2—CH_3)); 2.30—1.8 (m, $2xH_2C$—(CH_3)); 1.66 (d, H_3—C (3')); 1.33 (t, H_3C—(CH_2)).

Elemental analysis for $C_{17}H_{23}N_5O_5$

Calc. (%):	C 54.11,	H 6.1,	N 18.56
Found (%):	C 54.09,	H 6.99,	N 18.99

B) Preparation of 1',2'-dibutyryl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin

50 mg of PtO_2 were stirred with pure 25 ml of acetic acid at room temperature in the atmosphere of hydrogen for about 10 minutes until the catalyst was completely reduced and saturated with hydrogen. 500 mg of 1',2'-dibutyryl-L-biopterin were added. Dibutyryl-L-biopterin was not soluble in acetic acid and a suspension was obtained. It became a solution while a tetrahydro-derivative was produced by means of hydrogenation with stirring. The clear solution was obtained after 5 hours. The obtained solution was filtered from the catalyst and cooled in a freezing mixture (ice and salt). After freezing, a solution of 1 ml of 12 N HCl, 9 ml of methanol and 300 ml of ether was added and the whole flask was allowed to come to room temperature. The insoluble dibutyryl-tetrahydro-L-biopterin·2HCl was filtered, washed with ethanol and then ether until became free from HCl and dried in vacuo (0.0133 mbar) at 60°C for 16 hours for the elimination of the solvents. Yield: about quantitative.

The obtained 1',2'-dibutyryl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin was analyzed by means of ^1H—NMR (1N DCl).

The ^1H—NMR-spectrum chart is shown in Fig. 3: ^1H—NMR-spectrum analysis (the values of the chemical shifts are expressed in δ): (recorded in $CDCl_3$).

4.33—3.66 (m, H_2—C (7), H—C (6); H—C (1'); H—C (2')); 2.66 (m, $2xH_2C$—(CH_2—CH_3)); 1.90 (m, $2xH_2C$—(CH_3)); 1.60 (m, H_3—C (3')); 1.20 (t, H_3—C—(CH_2)).

Fig. 3 shows distinctly that the obtained compound was a diastereomeric mixture of 1',2'-dibutyryl-

(6R)-5',6',7',8'-tetrahydro-L-biopterin and 1',2-dibutyryl-(6S)-5',6',7',8'-tetrahydro-L-biopterin in a proportion of about 1:1.

Example 2

A) One pot synthesis of 1',2'-diacetyl-L-biopterin

A suspension of 14 g (42.1 mmol) of diethylsulfonyl-L-rhamnose in 120 ml of water was treated with 4N NH_4OH while stirring until the pH of the solution reached 9 to 10. After 14 hours-standing with stirring from time to time, the precipitate of diethylsulfonylmethane was filtered off and the filtrate was dried in vacuo at 40°C. The residue was dissolved in 80 ml of absolute methanol. After adding 5 g (46 mmol) of pure phenylhydrazine, the solution was kept at room temperature for 1 hour and then dried in vacuo at 40°C. The residue was washed two or three times with ether (50 ml each time) and dried. The dried residue was dissolved in 35 ml of pyridine and the solution was cooled. To the cooled solution of 0—5°C 35 ml of acetic anhydride were slowly added. After adding completely, the reaction mixture was allowed to stand in the ice-bath for 10 min and then it was kept at room temperature for 5 hours. After 200 ml of methanol were added to the solution, the solution was kept at room temperature for 10—15 hours (overnight). A solution of 1.0 g of sodium dithionite ($Na_2S_2O_4$) and 12.5 g of sodium acetate·$3H_2O$ dissolved in 300 ml of water, and a suspension of 12.0 g of 6-hydroxy-2,4,5-triamino-pyrimidine-sulfate·H_2O in 500 ml of water were then added successively to the methanol/pyridine solution. The whole reaction mixture was closed under nitrogen and stirred at 35—40°C for 20 hours to give an homogeneous, reddish brown solution. In the resulting solution the obtained tetrahydrobiopterin derivative was oxidized by adding 25 g of iodine dissolved in 300 ml of methanol. The excess of iodine, if any, was removed with sodium thiosulfate. If no excess was present, a small quantity of iodine solution was added in order to complete the oxidation of the tetrahydrobiopterin derivative. During the oxidation a fine brown crystalline precipitate was obtained.

The resulting oxidized solution was then concentrated to about 100 ml and filtered after cooling in the refrigerator for some hours. The filtered crude diacetylbiopterin was washed with 50 ml of cold water, 100 ml of cold ethanol, 100 ml of ether and dried. The dried residue was dissolved in about 1200 ml of boiling water and discolored with about 0.5 g of active charcoal. The solution was filtered while heating, the charcoal bed was washed with 50 ml of boiling water, the filtrate was allowed to cool to room temperature and then kept to 0—5°C for 10 hours.

The crystalline diacetyl-L-biopterin was filtered, washed twice with 50 ml of ethanol, then ether and dried to give 8.1 g of crystalline 1',2'-diacetyl-L-biopterin. Yield: 60%.

B) Preparation of 1',2'-diacetyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin

After reducing 350 mg of PtO_2 by a usual method with H_2 in 50 ml of trifluoroacetic acid, 1 g of pure 1',2'-diacetyl-L-biopterin was added to the obtained suspension. Then, H_2 was passed into the suspension. After 40 min the rate of H_2-uptake was slowed down, and the H_2-uptake was stopped after 45 min. The catalyst was quickly filtered and a colorless filtrate was frozen in liquid N_2. A cold mixture of 20 ml of ether was added thereto. While melting the frozen solution at room temperature, 1',2'-diacetyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin·2HCl was separated as a white powder. The powder was centrifuged, washed with acetonitrile and ether, dried over KOH in desiccator, and then dried under reduced pressure (0.0133 mbar) at 60°C for 15 hours. Yield: 1.1 g.

The obtained 1',2'-diacetyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin was analysed by means of ^1H—NMR (d_5-pyridin) and ^{13}C—NMR (D_2O).

The ^1H—NMR-spectrum charts are shown in Fig. 1 and Fig. 2.

Fig. 1 and Fig. 2 showed distinctly that the obtained compound was a diastereomeric mixture of 1',2'-diacetyl-(6R)-5,6,7,8-tetrahydro-L-biopterin and 1',2'-diacetyl-(6S)-5,6,7,8-tetrahydro-L-biopterin in a proportion of about 1:1.

Example 3

A) One pot synthesis of L-biopterin

A suspension of 14 g (42.1 mmol) diethylsulfonyl-L-rhamnose in 120 ml of water was treated with 4N NH_4OH while stirring until the pH of the solution reached 9 to 10. After 14 hours standing with stirring from time to time, the precipitate of diethylsulfonylmethane was filtered off and the filtrate was dried in vacuo at 40°C. The residue was dissolved in 80 ml of absolute methanol. After adding 5 g (46 mmol) of pure phenylhydrazine, the solution was kept at room temperature for 1 hour and then dried in vacuo at 40°C. The residue was washed two or three times with ether (50 ml each time) and dried. The dried residue was dissolved in 35 ml of pyridin and the solution was cooled. To the ice-cooled solution of 0—5°C 35 ml of acetic anhydride were slowly added. After adding completely, the reaction mixture was allowed to stand in the ice-bath for 10 min and then it was kept at room temperature for 5 hours. After 200 ml of methanol were added to the solution, the solution was kept at room temperature for 10—15 hours (overnight). A solution of 1.0 g of sodium dithionite ($Na_2S_2O_4$) and 12.5 g of sodium acetate·$3H_2O$ dissolved in 300 ml of water, and a suspension of 12.0 g of 6-hydroxy-2,4,5-triamino-pyrimidine-sulfate H_2O in 500 ml of water were then added successively to the methanol/pyridine solution. The whole reaction mixture was closed under nitrogen and stirred at 35—40°C for 20 hours to give an homogeneous, reddish brown solution. In the resulting solution the obtained tetrahydrobiopterin derivative was oxidized by adding 25 g of iodine

dissolved in 300 ml of methanol. The excess of iodine, if any, was removed with sodium thiosulfate. If no excess was present, a small quantity of iodine solution was added in order to complete the oxidation of the tetrahydrobiopterin derivative. During the oxidation a fine brown crystalline precipitate was obtained.

The resulting oxidized solution was concentrated in vacuo to about 100 ml 150 ml of ethanol and 250 ml of 14N NH_4OH were added to the concentrate. The resulting mixture was kept at 50°C for one hour. The deacetylated biopterin solution was then evaporated to dryness in vacuo at 40°C, the residue was taken up with 100 ml of methanol and filtered.

The crude biopterin was washed with 50 ml of cold water and 200 ml of ethanol. And then, without drying, it was dissolved in 1400 ml of boiling water with the smallest quantity of active charcoal to discolorize it. After hot filtration, the solution was allowed to cool to room temperature and then kept at 5°C for 10 hours. The crystalline biopterin was filtered, washed with cold water, ethanol, ether and then dried in vacuo (0.0133 mbar) at 40°C for 14 hours until became free from solvents to give 6 g of L-biopterin. Yield: 60%.

B) Preparation of (6R,S)-5,6,7,8-tetrahydro-L-biopterin

10 mg of PtO_2 were stirred with 30 ml of pure acetic acid at room temperature in the atmosphere of hydrogen for about 10 minutes until the catalyst was completely reduced and saturated with hydrogen. 500 mg of pure L-biopterin were then added. The L-biopterin was not soluble in acetic acid and a suspension was obtained. It was stirred in the atmosphere of hydrogen until all the L-biopterin became into solution. It took about 5 hours. The obtained solution was then filtered from the catalyst and the filtrate was cooled in a freezing mixture until it solidified. Then a solution of 9 ml of methanol, 90 ml of ether and 1 ml of 12N HCl was added to the solidified mixture and the whole flask was allowed to come to room temperature. The precipitated insoluble tetrahydrobiopterin·2HCl was filtered and washed with ethanol and then ether until became free from excess HCl and dried under reduced pressure. The crude tetrahydrobiopterin was crystallized with acetic acid. For the elimination of both solvents it was further dried in vacuo (0.0133 mbar) at 60°C for 16 hours. Yield: about quantitative.

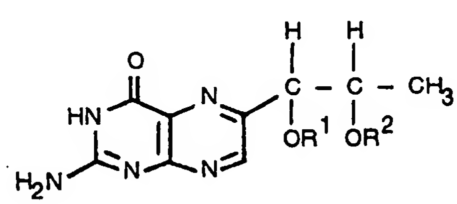
The following Table 1 shows the different times which are required for the complete catalytic reduction of L-biopterin by using several varieties of amount of biopterin, catalyst or acetic acid.

TABLE 1

Amount of biopterin (mg.)	Amount of catalyst (mg.)	Amount of solvent (Acetic acid) (ml.)	Time for complete reduction (hours)
100	40	6	1.5
100	20	6	1.5
100	10	6	2.0
200	10	12	3.0
300	10	18	4.0
400	10	24	4.5
500	10	30	5.0
1000	20	60	8.0

Claims

1. A process for preparing a 1',2'-diacyl-L-biopterin having the general formula (II):



(II)

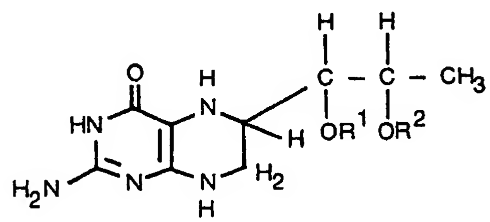
wherein R^1 and R^2 are the same or different and each is an acyl group by converting L-rhamnose to an acyl derivative of 5-deoxy-L-arabinose-phenylhydrazone through a 1,1-dialkylsulfonyl-L-rhamnose, reacting the obtained acyl derivative with 2,4,5-triamino-6-hydroxy-pyrimidine and then oxidizing the product with an oxidizing agent, characterised in that the reactions from the 1,1-dialkylsulfonyl-L-rhamnose to the 1',2'-diacyl-L-biopterin are carried out without isolating the intermediate products.

2. The process of claim 1, characterised in that the alkyl group of said 1,1-dialkylsulfonyl-L-rhamnose is a linear or branched alkyl group having 1 to 3 carbon atoms.

3. The process of claim 1, characterised in that said 1,1-dialkylsulfonyl-L-rhamnose is 1,1-diethylsulfonyl-L-rhamnose.

4. The process of claim 1, characterised in that said 1,1-dialkylsulfonyl-L-rhamnose is 1,1-dimethylsulfonyl-L-rhamnose.

5. The process of claim 1 for preparing a 1',2'-diacyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterin having the general formula (I):



(I)

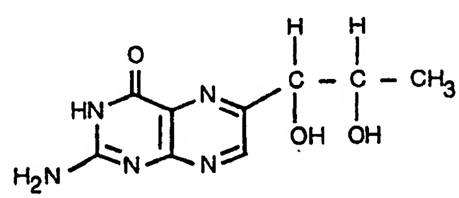
wherein R^1 and R^2 are as defined in claim 1, characterised in that the obtained 1',2'-diacyl-L-biopterin is catalytically hydrogenated.

6. The process of claim 5, characterised in that the alkyl group of said 1,1-dialkylsulfonyl-L-rhamnose is the methyl group.

7. The process of claim 5, characterised in that the alkyl group of said 1,1-dialkylsulfonyl-L-rhamnose is the ethyl group.

8. The process of claim 5, characterised in that said solvent does not dissolve the compound of formula (II), but dissolves the compound of the formula (I).

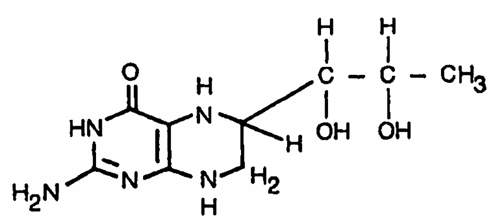
9. The process of claim 1 for preparing biopterin having the formula (II'):



(II')

characterised in that the resultant 1',2'-diacyl-L-biopterin is deacylated.

10. The process of claim 1 for preparing a (6R,S)-5,6,7,8-tetrahydro-L-biopterin having the formula (I'):

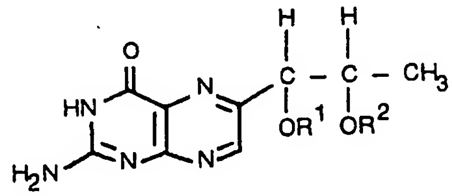


(I')

characterised in that the resultant 1',2'-diacyl-L-biopterin is deacylated and catalytically hydrogenated.

Patentansprüche

1. Verfahren zur Herstellung eines 1',2'-Diacyl-L-biopterins der allgemeinen Formel (II):



(II)

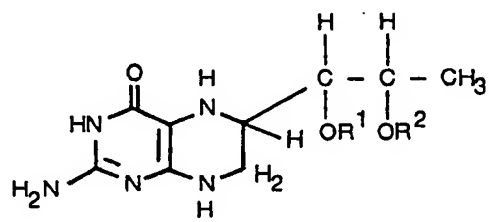
worin R^1 und R^2 gleich oder verschieden sind und jeweils eine Acylgruppe bedeuten, durch Umwandlung von L-Rhamnose in ein Acylderivat von 5-Deoxy-L-arabinose-phenylhydrazon über eine 1,1-Dialkylsulfonyl-L-rhamnose, Umsetzung des erhaltenen Acylderivats mit 2,4,5-Triamino-6-hydroxy-pyrimidin und anschließendes Oxidieren des Produkts mit einem Oxidationsmittel, dadurch gekennzeichnet, daß die Reaktionen von der 1,1-Dialkylsulfonyl-L-rhamnose bis zu dem 1',2'-Diacyl-L-biopterin ohne Isolierender Zwischenprodukte durchgeführt werden.

2. Verfahren nach Anspruch 1, dadurch gekennzeichnet, daß die Alkylgruppe der 1,1-Dialkylsulfonyl-L-rhamnose eine lineare oder verzweigte Alkylgruppe mit 1 bis 3 Kohlenstoffatomen ist.

3. Verfahren nach Anspruch 1, dadurch gekennzeichnet, daß die 1,1-Dialkylsulfonyl-L-rhamnose 1,1-Diethylsulfonyl-L-rhamnose ist.

4. Verfahren nach Anspruch 1, dadurch gekennzeichnet, daß die 1,1-Dialkylsulfonyl-L-rhamnose 1,1-Dimethylsulfonyl-L-rhamnose ist.

5. Verfahren nach Anspruch 1 zur Herstellung eines 1',2'-Diacyl-(6R,S)-5,6,7,8-tetrahydro-L-biopterins der allgemeinen Formel (I)



(I)

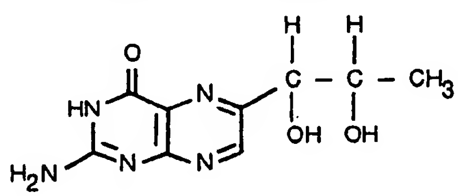
worin R^1 und R^2 wie in Anspruch 1 definiert sind, dadurch gekennzeichnet, daß das erhaltene 1',2'-Diacyl-L-biopterin katalytisch hydriert wird.

6. Verfahren nach Anspruch 5, dadurch gekennzeichnet, daß die Alkylgruppe der 1,1-Dialkylsulfonyl-L-rhamnose die Methylgruppe ist.

7. Verfahren nach Anspruch 5, dadurch gekennzeichnet, daß die Alkylgruppe der 1,1-Dialkylsulfonyl-L-rhamnose die Ethylgruppe ist.

8. Verfahren nach Anspruch 5, dadurch gekennzeichnet, daß das Lösungsmittel die Verbindung der Formel (II) nicht löst, jedoch die Verbindung der Formel (I) löst.

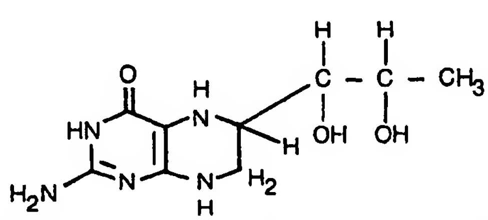
9. Verfahren nach Anspruch 1, zur Herstellung von Biopterin der Formel (II'):



(II')

dadurch gekennzeichnet, daß das resultierende 1',2'-Diacyl-L-biopterin deacyliert wird.

10. Verfahren nach Anspruch 1 zur Herstellung eines (6R,S)-5,6,7,8-Tetrahydro-L-biopterins der Formel (I'):

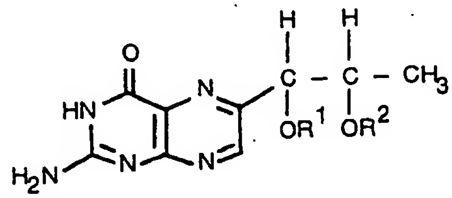


(I')

dadurch gekennzeichnet, daß das resulierende 1',2'-Diacyl-L-biopterin deacyliert und katalytisch hydriert wird.

Revendications

1. Un procédé pour préparer une 1',2'-diacyl-L-bioptérine répondant à la formule générale II:



(II)

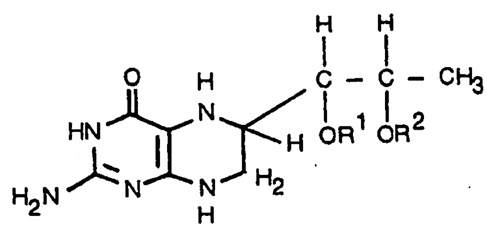
dans laquelle R^1 et R^2 sont semblables ou différents et sont chacun un groupe acyle, par transformation du L-rhamnose en un dérivé acylé de la 5-désoxy-L-arabinose-phénylhydrazone par l'intermédiaire d'un 1,1-dialkylsulfonyl-L-rhamnose, réaction du dérivé acylé obtenu avec la 2,4,5-triamino-6-hydroxypyrimidine puis oxydation du produit avec un agent oxydant, caractérisé en ce que les réactions du 1,1-dialkylsulfonyl-L-rhamnose à la 1',2'-diacyl-L-bioptérine sont effectuées sans isolement des produits intermédiaires.

2. Le procédé de la revendication 1, caractérisé en ce que le groupe alkyle dudit 1,1-dialkylsulfonyl-L-rhamnose est un groupe alkyle linéaire ou ramifié ayant 1 à 3 atomes de carbone.

3. Le procédé de la revendication 1, caractérisé en ce que ledit 1,1-dialkylsulfonyl-L-rhamnose est le 1,1-diéthylsulfonyl-L-rhamnose.

4. Le procédé de la revendication 1, caractérisé en ce que ledit 1,1-dialkylsulfonyl-L-rhamnose est le 1,1-diméthylsulfonyl-L-rhamnose.

5. Le procédé de la revendication 1 pour la préparation d'une 1',2'-diacyl-(6R,S)-5,6,7,8-tétrahydro-L-bioptérine répondant à la formule générale I:



(I)

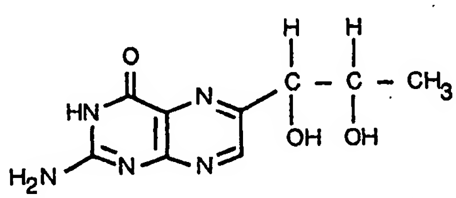
dans laquelle R^1 et R^2 sont comme définis dans la revendication 1, caractérisé en ce que la 1',2'-diacyl-L-bioptérine obtenue est hydrogénée catalytiquement.

6. Le procédé de la revendication 5, caractérisé en ce que le groupe alkyle dudit 1,1-dialkylsulfonyl-L-rhamnose est le groupe méthyle.

7. Le procédé de la revendication 5, caractérisé en ce que le groupe alkyle dudit 1,1-dialkylsulfonyl-L-rhamnose est le groupe éthyle.

8. Le procédé de la revendication 5, caractérisé en ce que ledit solvant ne dissout pas le composé de formule II mais dissout le composé de formule I.

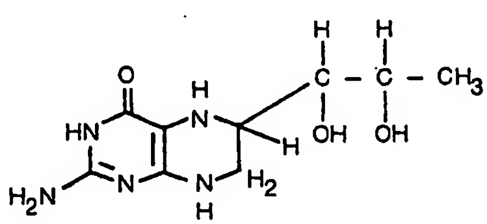
9. Le procédé de la revendication 1 pour la préparation de la bioptéridine répondant à la formule II':



(II')

caractérisé en ce que la 1',2'-diacyl-L-bioptérine obtenue est désacylée.

10. Le procédé de la revendication 1, pour préparer une (6R,S)-5,6,7,8-tétrahydro-L-bioptérine répondant à la formule I':



(I')

caractérisé en ce que la 1',2'-diacyl-L-bioptérine obtenue est désacylée et hydrogénée catalytiquement.

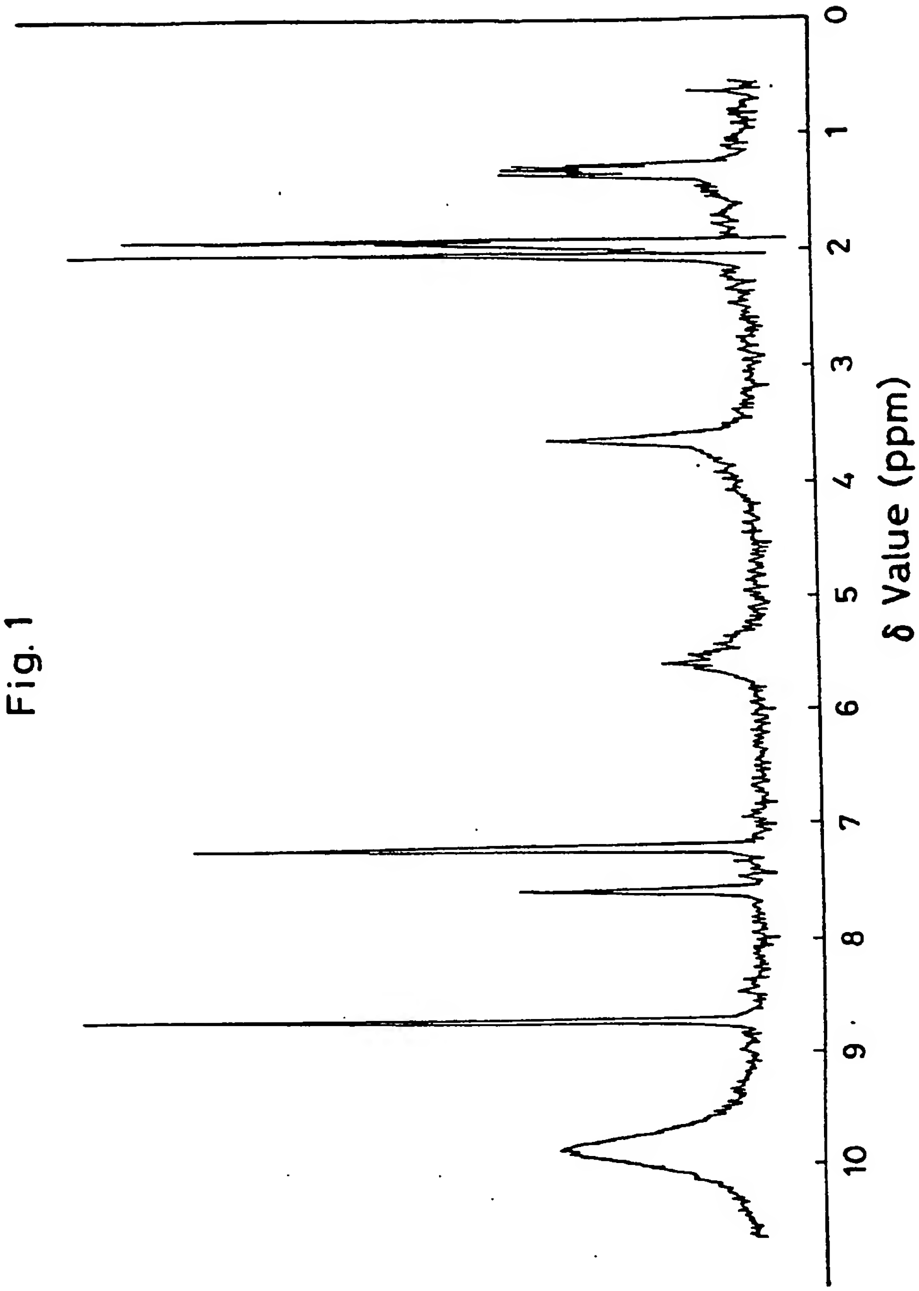


Fig. 1

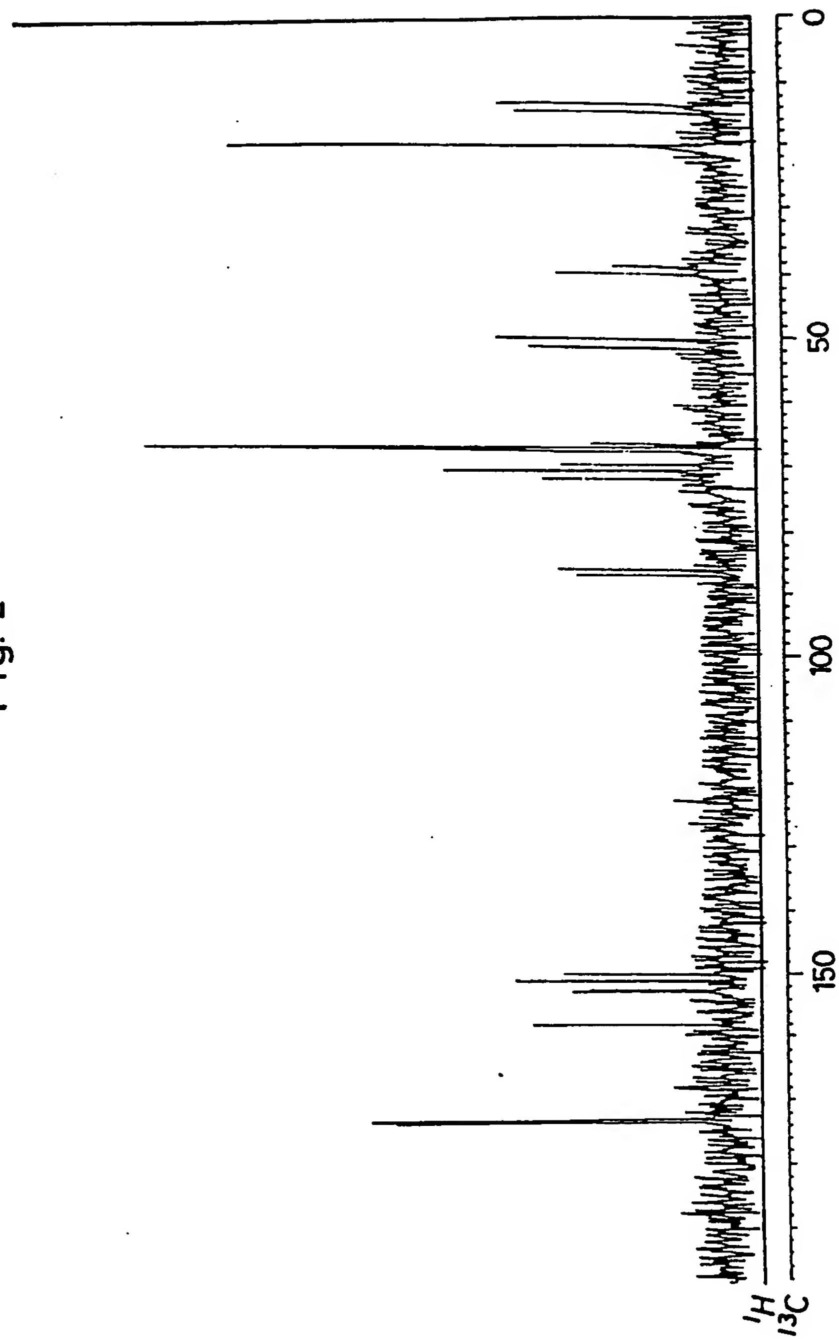


Fig. 2

Fig. 3

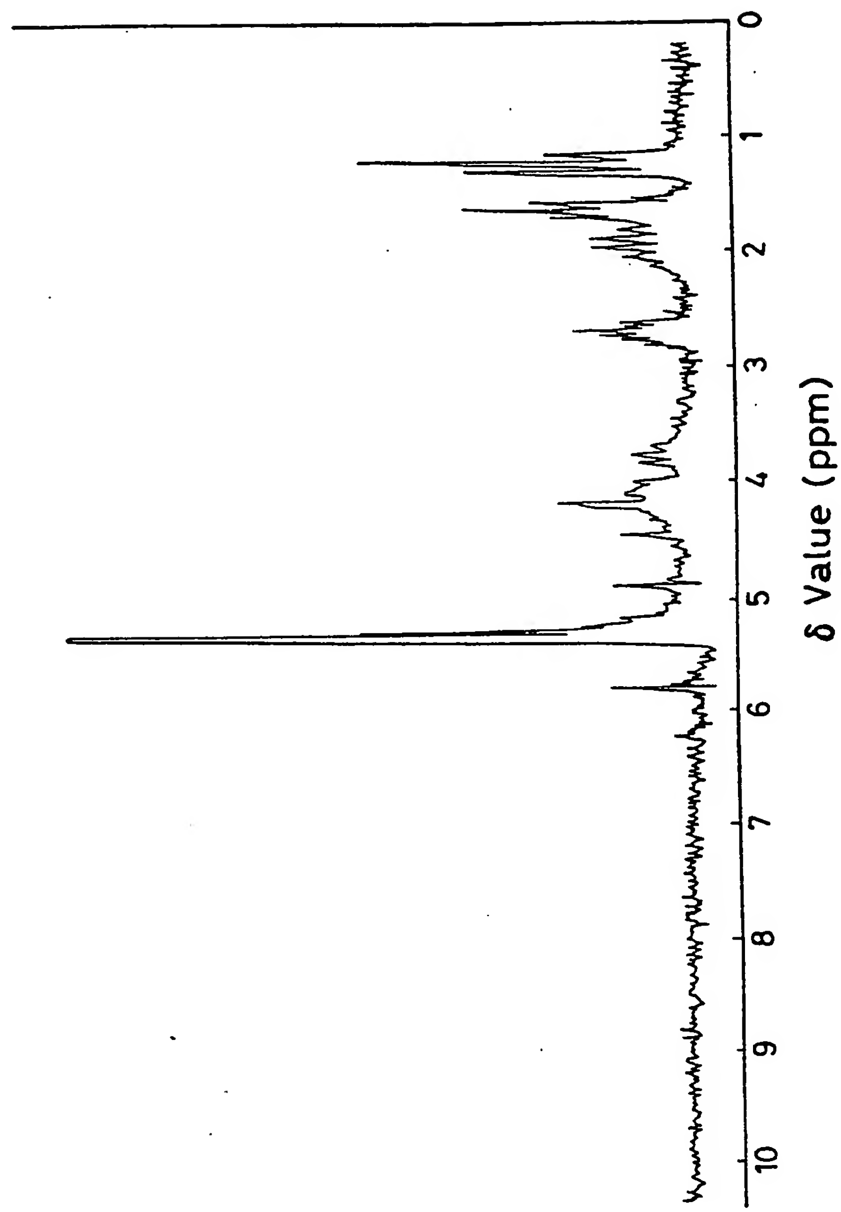


Fig. 4